Improvement of growth and yield of several varieties of soybean following paddy rice using *Rhizobium* biofertilizer with reduced NPK doses

W Wangiyana1,2,*, D F Arianti1 and I K Ngawit1

1 Faculty of Agriculture, University of Mataram, Mataram, Lombok, Indonesia
2 Postgraduate Program, University of Mataram, Mataram, Indonesia
3 Graduate of Faculty of Agriculture, University of Mataram, Mataram, Indonesia

*Email: w.wangiyana@unram.ac.id

**Abstract.** In paddy fields, soybean is generally planted following harvest of paddy rice crops, and the average yield achieved by farmers is still low. This study aimed to examine the effect of applying *Rhizobium* biofertilizer with reduced NPK fertilizer dose on growth and yield of soybean grown following flooded rice crop. The experiment, conducted on farmers’ paddy fields in East Lombok (Indonesia) from August to November 2020, was arranged according to Randomized Block Design, with two treatment factors, namely soybean varieties (Anjasmoro, Grobogan, Biosoy-2) and biofertilizers (P0= without biofertilizer, P1= 50% NPK dose + *Rhizobium* biofertilizer). The results showed that application of *Rhizobium* biofertilizer did not significantly increase growth and yield of soybean except for weight of 100 grains, but the significant interaction effect on trifoliate number, pod number, unfilled pod number, weight of 100 grains, harvest index, and grain yield indicated that grain yields of Grobogan (2.03 tons/ha) and Biosoy-2 (2.31 tons/ha) were significantly higher with biofertilizer application while that of Anjasmoro variety (2.83 tons/ha) was higher under recommended NPK fertilization doses. It was also concluded that the NPK fertilizer dose can be reduced by 50% of the recommended dose if it is accompanied by application of *Rhizobium* biofertilizer.

1. **Introduction**

There are many important crops in the world, which produce high quality seeds for use as food materials. One of them is soybean [*Glycine max* (L) Merr.], which produces seeds with high content of protein (34 - 57%), and about 66% of the world’s protein meal is from soybean seeds [1]. Due to its high protein content, soybean seed can be a cheaper source of proteins for human health compared with meat. Based on the total area ([https://www.bps.go.id/indicator/53/21/1/luas-panen.html](https://www.bps.go.id/indicator/53/21/1/luas-panen.html)), soybean in Indonesia is the most important food crop belonging to the family Leguminosae (or legume crops). Unfortunately, due to its relatively very low productivity averaging 1,568 kg/ha and relatively low production areas ([https://www.bps.go.id/indicator/53/22/1/produktivitas.html](https://www.bps.go.id/indicator/53/22/1/produktivitas.html)), while the domestic need for soybean has been constantly very high, then the total soybean production has not been able to meet the need for it. The total soybean production of 963,183 ton in 2015 ([https://www.bps.go.id/indicator/53/23/1/produksi.html](https://www.bps.go.id/indicator/53/23/1/produksi.html)) was far below the domestic need for it, so
that Indonesia imported soybean seeds, in which the volume of import was much higher than the domestic production. The total import volume was as high as 2.5 million tons in 2021 [2].

To reduce import, the domestic production of soybean needs to be increased either by increasing the total area harvested or the productivity of soybean or both because the national average productivity of soybean is still very low. The low productivity of soybean normally achieved by the farmers in Indonesia could be due to the less optimum conditions of the growing environment of soybean crop, which is mostly grown during the dry season after rice crops. In fact, based the soybean description, some national soybean varieties have a high grain yield potential, such as Grobogan (3.4 ton/ha), and Kipas Merah Bireun (3.5 ton/ha) [3]. Based on the research results by Wangiyana and Farida [4], application of Rhizobium and mycorrhiza biofertilizers on soybean direct seeded after harvest of rainfed paddy rice produced much higher total grain yield than the same soybean varieties fertilized with the recommended dose of NPK fertilizer. Different techniques of growing rice crop also reported to have significant effects on yield of soybean plants grown after the rice crop [5]. Direct-seeding soybean following harvest of aerobic red rice grown on raised-bed also resulted in significantly higher grain yield than direct-seeding it following harvest of normally flooded rice crop [6].

Rice in an irrigated rice growing areas is mostly grown under a conventional technique in which rice crop is mostly flooded during its growing period, and there is a tendency for the farmers to grow all the time whenever irrigation water is available. This is also the case in the rice growing areas in Masbagik (East Lombok) where the experiment in this study was done. One of the obstacles in an effort to obtain reasonably high grain yield of soybean grown in rotation with paddy rice is the low content of AMF (arbuscular mycorrhizal fungi) propagules in the soil after the paddy rice [7, 8]. In addition, rice plants normally acidify their rhizosphere when flooded, which resulted in lower pH of the soils after harvest of paddy rice [9]. These were thought to have a bad impact on the performance and yield of soybean plants grown after harvest of paddy rice [8]. Therefore, the objective of this study was to examine the effect of application of Rhizobium biofertilizer accompanied with reduced NPK doses on growth and yield of several varieties of soybean grown following paddy rice in Masbagiq (East Lombok, Indonesia).

2. Materials and Methods
The experiment reported in this study was conducted in a farmer’s paddy field in Masbagiq District (East Lombok, Indonesia) from August to November 2020, after harvest of a second rice crop. The biofertilizer used was in the form of powdery inoculant containing Rhizobium sp, Azotobacter sp, Trichoderma harzianum, Aspergilus niger and Pseudomonas fluorescens, under the trademark “Soyaku”, while the NPK fertilizer used was Phonska Plus (NPK 15-15-15 plus Zn 2000 ppm and 9% S).

2.1. Treatments and experimental design
There were two treatment factors tested in this experiment, namely soybean varieties (V1= Anjasmoro, V2= Grobogan, V3= Biosoy-2) and biofertilizer application (P0 = without biofertilizer (fertilization was only with the recommended dose of NPK), P1 = application of Rhizobium biofertilizer accompanied with NPK fertilizer of only 50% of the recommended dose). These resulted in six treatment combinations, which were arranged according to the Randomized Complete Block Design, and each combination was made in three blocks (replications). The NPK used was Phonska Plus from Gresik Petrochemicals in the form of white granules containing 15% N, 15% P, 15% K, 9% S and 2000 ppm Zn with a recommended dose of 250 kg/ha.

2.2. Implementation of the experiment
After the second rice crop was harvested, the land preparation was done without tillage, by only making furrows of 30 cm width surrounding planting beds of 175 cm length and 100 cm width, with
the furrow depth of 20-25 cm. After covering the planting beds with plastic mulch, to reduce weed growth, soybean seeds were dibbled with a plant spacing of 35 cm x 20 cm. For the treatments with biofertilizer application, soybean seeds were coated with the *Rhizobium* biofertilizer powder prior to dibbling into the planting holes, by first wetting the seeds.

NPK fertilization was done using Phonska Plus, with the recommended dose of 250 kg/ha, which was split into two applications of 125 kg/ha each. For the P0 treatment (without biofertilizer), the first NPK dose was applied at seeding and the second split was applied at 30 days after seeding (DAS), by dibbling the NPK fertilizer about 5 cm beside the planting hole at 5 cm depth. For the P1 treatment (with biofertilizer) the NPK dose was reduced to only 50% (125 kg/ha) which was applied at 30 DAS as described for the P0 treatment. Pest control was done using the insecticide Regent 50 SC, especially for controlling the pod borers. Planting was carried out on land without conventional post-rice tillage with a spacing of 35x20 cm by sampling 4 clumps of plant samples in one experimental plot. Harvest of the plant samples was done after 80 DAS depending on the variety, i.e. when most pods were already showing brown colour of the pod skin.

### 2.3. Observation variables and data analysis

The observation variables were plant height and trifoliate number (measured every week which was started two weeks after seeding) but the data analyzed were the results of measurements at the peak of flowering i.e. at 35 days after seeding (DAS) and seed-filling stage (56 DAS), AGR (average growth rate) of plant height, pod number at 56 DAS, and yield components measured after harvest (dry stover weight, filled pod number, weight of 100 dry grains, harvest index, grain yield per clump and its conversion to per ha as the potential yield). The data were analyzed with Analysis of Variance (ANOVA) and Tukey’s HSD at 5% level of significance, using CoStat for Windows ver. 6.303.

### 3. Results and discussion

The results of ANOVA and HSD test for growth related variables (Table 1) show that there was a significant interaction effect on the trifoliate number at 35 DAS, and it was also significantly different among the varieties of soybean but it was higher under full NPK fertilization than under application of biofertilizer with 50% dose of NPK fertilizer.

<table>
<thead>
<tr>
<th>Treatments</th>
<th>Plant height 35 DAS (cm)</th>
<th>Plant height 56 DAS (cm)</th>
<th>AGR of plant height (cm/day)</th>
<th>Trifoliate number 35 DAS</th>
<th>Trifoliate number 56 DAS</th>
<th>Dry stover weight (g/clump)</th>
</tr>
</thead>
<tbody>
<tr>
<td>V1: Anjasmoror</td>
<td>49.17 a</td>
<td>63.85 a</td>
<td>0.60 a</td>
<td>9.84 c</td>
<td>16.86 a</td>
<td>35.16 a</td>
</tr>
<tr>
<td>V2: Grobogans</td>
<td>38.42 b</td>
<td>48.75 b</td>
<td>0.44 ab</td>
<td>11.63 b</td>
<td>16.33 a</td>
<td>36.04 a</td>
</tr>
<tr>
<td>V3: Biosoy-2</td>
<td>34.08 b</td>
<td>44.21 b</td>
<td>0.37 b</td>
<td>14.04 a</td>
<td>18.06 a</td>
<td>41.05 a</td>
</tr>
<tr>
<td>HSD 0.05</td>
<td>5.06</td>
<td>7.54</td>
<td>0.23</td>
<td>1.25</td>
<td>ns</td>
<td>ns</td>
</tr>
<tr>
<td>P0: NPK 100%</td>
<td>41.46 a</td>
<td>54.43 a</td>
<td>0.50 a</td>
<td>12.56 a</td>
<td>17.63 a</td>
<td>42.28 a</td>
</tr>
<tr>
<td>P1: Bio+NPK50%</td>
<td>39.65 a</td>
<td>50.11 a</td>
<td>0.43 a</td>
<td>11.11 b</td>
<td>16.54 a</td>
<td>32.55 b</td>
</tr>
<tr>
<td>HSD 0.05</td>
<td>ns</td>
<td>ns</td>
<td>ns</td>
<td>0.83</td>
<td>ns</td>
<td>6.80</td>
</tr>
</tbody>
</table>

Interaction effect ns ns ns ** ns ns

1) The same letters indicate non-significant differences between levels of each treatment factor

In general, growth variables were not significantly reduced by the 50% reduction in the NPK fertilizer dose, except for trifoliate number and dry stover weight per plant, which were significantly reduced (Table 1). However, the patterns of interaction show that the significant reduction in the
Trifoliate number at 35 DAS was observed only in Grobogan variety, while on the other two varieties it was not significantly reduced (Fig. 1). However, this was not expressed in the grain yield. It appears that the reduction in trifoliate number at 35 DAS was not exactly manifested in the grain yield per plant, in which the reduction in trifoliate number at 35 DAS only reduced grain yield of Anjasmoro variety (V1) due to the 50% reduction in NPK dose, while in Grobogan variety (V2), the lower trifoliate number at 35 DAS in P1 treatment (Fig. 1) was associated with higher grain yield (Fig. 2). Similarly, grain yield of Biosoy-2 variety (V3) was also significantly higher under P1 than P0 treatments (Fig. 2).

In relation to yield components of soybean, 50% reduction in NPK dose accompanied with application of *Rhizobium* biofertilizer was not significantly reduced yield components, including numbers of pods at 56 DAS (which was the seed-filling stage). Other yield components such as numbers of filled pod at harvest, weight of 100 grains, grain yield, potential yield, and harvest index, were in fact increased by application of *Rhizobium* biofertilizer but with reduced NPK dose although the increase was significant only in weight of 100 grains and harvest index (Table 2).

![Fig. 1. Trifoliate number per plant at 35 DAS (Mean ± SE) as affected by the P*V interactions](image1.png)

![Fig. 2. The potential grain yield in ton/ha (Mean ± SE) as affected by the P*V interactions](image2.png)

<table>
<thead>
<tr>
<th>Treatments</th>
<th>Pod number 56 DAS</th>
<th>Filled pod number</th>
<th>Weight of 100 grains (g)</th>
<th>Grain yield (g/clump)</th>
<th>Potential grain yield (ton/ha)</th>
<th>Harvest index (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>V1: Anjasmoro</td>
<td>29.29 a</td>
<td>25.15 a</td>
<td>24.35 c</td>
<td>17.52 a</td>
<td>2.50 a</td>
<td>33.51 a</td>
</tr>
<tr>
<td>V2: Grobogan</td>
<td>23.21 b</td>
<td>17.17 b</td>
<td>28.82 b</td>
<td>12.87 b</td>
<td>1.84 b</td>
<td>27.82 b</td>
</tr>
<tr>
<td>V3: Biosoy-2</td>
<td>24.61 b</td>
<td>22.44 ab</td>
<td>38.93 a</td>
<td>14.15 b</td>
<td>2.02 b</td>
<td>25.74 b</td>
</tr>
<tr>
<td>HSD 0.05</td>
<td>4.37</td>
<td>6.55</td>
<td>2.71</td>
<td>2.31</td>
<td>0.33</td>
<td>4.53</td>
</tr>
<tr>
<td>P0: NPK 100%</td>
<td>25.72 a</td>
<td>20.74 a</td>
<td>28.79 b</td>
<td>14.47 a</td>
<td>2.07 a</td>
<td>25.72 b</td>
</tr>
<tr>
<td>P1: Bio+NPK50%</td>
<td>25.69 a</td>
<td>22.43 a</td>
<td>32.61 a</td>
<td>15.22 a</td>
<td>2.17 a</td>
<td>32.32 a</td>
</tr>
<tr>
<td>HSD 0.05</td>
<td>ns</td>
<td>ns</td>
<td>1.80</td>
<td>ns</td>
<td>ns</td>
<td>3.01</td>
</tr>
</tbody>
</table>

Interaction ns ns *** *** *** **

1) The same letters indicate non-significant differences between levels of each treatment factor
These two variables, i.e. weight of 100 grains and harvest index, seem to be the most influencing variables in increasing grain yield (Fig. 3 and Fig. 4). Although mean values of growth variables were reduced to the 50% reduction in NPK dose (Table 1), grain yield was increased by the NPK reduction accompanied with *Rhizobium* biofertilizer, especially Grobogan (V2) and Biosoy-2 (V3) varieties of soybean (Fig. 2). Regarding to the increase in grain yield, Fig. 3 & Fig. 4 show that grain yield increases were associated with an increase in the weight of 100 grains (Fig. 3) and harvest index (Fig. 4). Higher weight of 100 grains of a soybean variety due to a treatment factor such as *Rhizobium* biofertilizer application as shown by V2 and V3 (Fig. 3) could indicated that allocation of assimilates and nutrients to the grains was higher per grain in soybean plants supplied with *Rhizobium* biofertilizer than in those without biofertilizer application. This was supported by higher harvest index in V2 and V3 supplied with *Rhizobium* biofertilizer compared with no application of the biofertilizer, which means that the percentage of biomass partition to the grains in V2 and V3 soybean plants supplied with the biofertilizer was higher than in the varieties without application of the biofertilizer (Fig. 4). However, a regression analysis between weight of 100 grains and grain yield per clump does not show a significant relationship but regression analysis between harvest index and grain yield per clump shows a highly significant $R^2$ of 41.93% ($p<0.01$), with a regression equation is $Y$ (grain yield) = 5.9 + 0.31 $X$ ($p<0.01$). Previous study also reported that application of *Rhizobium* and mycorrhiza biofertilizers significantly increased weight of 100 grains, harvest index and grain yield per clump of Anjasmoro and Wilis varieties of soybean grown following a rainy season rice crop on vertisol land [4].

Based on the brochure of the “Soyaku” biofertilizer used in the field experiment in this study, this biofertilizer contains several species of microbes that can function as biofertilizer, namely *Rhizobium* sp, *Azotobacter* sp, *Trichoderma harzianum*, *Aspergillus niger* and *Pseudomonas fluorescens*. Therefore, in addition to containing microbes capable of performing biological nitrogen fixation (BNF) such as *Rhizobium* sp and *Azotobacter* sp, this biofertilizer also contains phosphate solubilizing microbes, such as *Aspergillus niger* and *Pseudomonas fluorescens*, that can make phosphates in the unavailable forms in the soil to become available for uptake by the soybean plants. $N_2$-fixing microbes are very important in increasing N content of the soil through N-rhizodeposition, which makes more nitrogen available for uptake by crops [10]. Similarly, phosphate solubilizing microbes are capable of transforming unavailable P, which is highly immobile in soil, to become available for uptake by plant roots, and they are keys for sustainable agriculture [11].

In relation to the main effect of the varieties used, most growth variables (Table 1) and all yield components (Table 2) were significantly different between varieties of soybean used in this study. The
most highly significant different between the varieties used was the weight of 100 grains. Among the varieties of soybean, Biosoy-2 variety produced the largest size of grains. In contrast, the harvest index was highest in Anjasmoro variety (Table 2). However, the weight of 100 grains, grain yield per clump, and harvest index showed a significant interaction effect between the treatment factors (Table 2). Based on this interaction effect on grain yield per clump or potential grain yield in ton/ha, it can be seen from Fig. 2 that in both Grobogan and Biosoy-2 varieties the potential grain yield was higher under application of the biofertilizer in spite of a 50% reduction of NPK dose than under the 100% NPK recommended dose. In contrast, the Anjasmoro variety produce higher grain yield under full NPK fertilization than under application of 50% NPK dose although with application of the biofertilizer. Therefore, it can be said that both Grobogan and Biosoy-2 are more responsive to application of the *Rhizobium* biofertilizer while Anjasmoro variety was more responsive to NPK fertilization.

4. Conclusion

It was concluded that application of *Rhizobium* biofertilizer had a significant effect only on few variables while differences in the varieties of soybean had significant effect on almost all variables. However, there were significant interaction effects between the treatment factors on trifoliate number at 35 DAS, weight of 100 grains, grain yield and harvest index. It can be concluded that both Grobogan and Biosoy-2 varieties are more responsive to application of the *Rhizobium* biofertilizer while Anjasmoro variety was more responsive to NPK fertilization. Therefore, in both Grobogan and Biosoy-2 varieties, the dose of NPK fertilizer can be reduced by 50% from the recommended dose if it is accompanied by application of *Rhizobium* biofertilizer.

5. References


